

-108-

WHAT IS CLAIMED IS:

1. A modified adenovirus fiber, comprising a modification to the fiber protein shaft, wherein the modification comprises a modification selected from among:
 - 5 a modification of a last full repeat;
 - a modification of a β -repeat corresponding to a third β -repeat and a modification of a last full repeat; and
 - a modification of one or both of a modification of a last full repeat and a modification of at least one amino acid in a contiguous sequence of
 - 10 amino acids corresponding to the amino acid sequence TTVT/S set forth in SEQ ID No. 44 in a third β -repeat, whereby binding of the fiber or of a viral particle containing such fiber to the Coxsackie-Adenovirus Receptor (CAR) is reduced compared to the unmodified fiber.
2. A modified adenovirus fiber, comprising a modification to the
- 15 fiber protein shaft, whereby binding of the modified fiber to Coxsackie-Adenovirus Receptor (CAR) is reduced or eliminated, wherein:
 - the unmodified fiber binds the Coxsackie-Adenovirus Receptor (CAR); and
 - 20 the modification comprises a modification of a repeat corresponding to the last full β -repeat, or the third β -repeat and the last full β -repeat of the shaft or one or both of the last full β -repeat and a portion of the third β -repeat that comprises the TTVT/S motif (SEQ ID No. 44) or a corresponding motif.
- 25 3. A modified fiber of claim 1 or 2, wherein the modified fiber binds to CAR with less than 50%, 40%, 30%, 20%, 10%, 5%, 1% of the binding affinity of the unmodified fiber.
4. A modified adenovirus fiber of any of claims 1-3, wherein the modified fiber is more rigid than the unmodified fiber.

-109-

5. A modified adenovirus fiber of any of claims 1-4, wherein the modification is a mutation, deletion, insertion or replacement of at least one amino acid in the fiber shaft repeat corresponding to the third repeat.
- 5 6. The modified adenovirus fiber of any of claims 1-5, wherein the unmodified fiber is a fiber of a serotype C adenovirus.
7. The modified adenovirus fiber of claim 6, wherein the serotype C adenovirus is Ad2 or Ad5.
8. The modified adenovirus fiber of any of claims 1-7, wherein
10 the modified fiber is more rigid and shorter than the unmodified fiber.
9. The modified adenovirus fiber of any of claims 1-8, wherein at least one amino acid in the contiguous sequence of amino acids corresponding to the amino acid sequence set forth in SEQ ID No. 42 or 43 is modified.
- 15 10. The modified adenovirus fiber of any of claims 1-6, wherein the third β -repeat is modified by replacing it with a corresponding repeat from a serotype D fiber shaft repeat sequence.
11. The modified adenovirus fiber of claim 10, wherein the serotype D adenovirus is selected from the group consisting of Ad8, Ad9,
20 Ad15, Ad19p and Ad37.
12. The modified adenovirus fiber of any of claims 1-6, wherein the third β -repeat is modified by replacing it with a corresponding repeat selected from the group consisting of SEQ ID NOS: 58, 66, 67 and 68.
13. The modified adenovirus fiber of any of claims 1-12, wherein
25 the modification is a mutation, deletion, insertion or replacement of at least one amino acid in a fiber shaft β -repeat corresponding to the last full β repeat and/or corresponding to the third β repeat.
14. The modified adenovirus fiber of claim 13, wherein the unmodified fiber is a serotype C adenovirus fiber.

-110-

15. The modified adenovirus fiber of claim 12, wherein the serotype C adenovirus is Ad2 or Ad5.

16. The modified adenovirus fiber of claim 15, wherein the modification is a modification of at least one amino acid in a contiguous
5 sequence of amino acids corresponding to those set forth in SEQ ID No. 46 or SEQ ID No. 47.

17. The modified adenovirus fiber of any of claims 1-15, wherein the modification comprises replacement of the last full β -repeat with a corresponding repeat sequence from a serotype D adenovirus fiber shaft.

10 18. The modified adenovirus fiber of claim 17, wherein the serotype D adenovirus is selected from the group consisting of Ad8, Ad9, Ad15, Ad19p and Ad37.

19. The modified adenovirus fiber of any of claims 13-18, wherein the modification is in the last full repeat; and the last full repeat
15 comprises a change of at least one amino acid in the repeat at contiguous amino acids corresponding to the amino acid sequence set forth in SEQ ID No. 49.

20. The modified adenovirus fiber of any of claims 13-18, wherein the modification is in the last full repeat; and the last full repeat
20 comprises a sequence of amino acids selected from the group consisting of SEQ ID NOS: 48, 59, 60 and 61.

21. The modified adenovirus fiber of any of claims 1-7, wherein a contiguous sequence of amino acids corresponding to the third repeat of the fiber shaft is deleted.

25 22. The modified adenovirus fiber of any of claims 1-7, wherein a contiguous sequence of amino acids corresponding to the last full repeat of the fiber shaft is deleted.

23. The modified adenovirus fiber of any of claims 1-7, wherein a contiguous sequence of amino acids corresponding to the third repeat

-111-

and the contiguous sequence of amino acids corresponding to the last full repeat are modified.

24. The modified adenovirus fiber of claim 22 or claim 23, wherein the modification is a mutation, deletion, insertion or replacement
5 of at least one amino acid in a fiber shaft repeat corresponding to the third repeat and/or the last full repeat.

25. The modified adenovirus fiber of claim 24, wherein the unmodified fiber shaft is from a serotype C adenovirus.

26. The modified adenovirus fiber of claim 25, wherein the
10 serotype C adenovirus is Ad2 or Ad5.

27. The modified adenovirus fiber of claim 25, wherein the modified repeats corresponding to the third repeat and the last full repeat are from a serotype D adenovirus.

28. The modified adenovirus fiber of claim 27, wherein the
15 serotype D adenovirus is selected from the group consisting of Ad8, Ad9, Ad15, Ad19p and Ad37.

29. The modified adenovirus fiber of claim 25, wherein the third repeat comprises a sequence selected from the group consisting of SEQ ID NOs. 58, 66, 67 and 68 and the last full repeat comprises an amino
20 acid sequence selected from the group consisting of SEQ ID NOs. 48, 59, 60 and 61.

30. The modified adenovirus fiber of claim 25, wherein the third repeat sequence is selected from a corresponding repeat sequence of a fiber protein from Ad8, Ad9, Ad15, Ad19p or Ad37; and the last full
25 repeat is selected from a corresponding repeat sequence of a fiber protein from Ad8, Ad9, Ad15, Ad19p or Ad37.

31. The modified adenovirus fiber of any of claims 1-30, wherein the modified adenovirus fiber further comprises at least one additional modification in the fiber protein, whereby the modified fiber binds to a

-112-

receptor other than CAR with greater affinity than the unmodified fiber binds to such receptor.

32. The modified adenovirus fiber of any of claims 1-30, wherein the modified adenovirus fiber further comprises at least one additional
5 modification in the fiber protein; and the modification is a modification in the fiber knob that further reduces or eliminates any binding of the modified fiber to CAR.

33. The modified adenovirus fiber of claim 31, wherein an additional modification is a modification of the Heparin Sulfate
10 Proteoglycans (HSP) binding site in the fiber shaft.

34. The modified adenovirus fiber of claim 31 or claim 32, wherein an additional modification is a modification in the fiber knob.

35. The modified fiber of any of claims 1-34, wherein the fiber is shortened or its flexibility is reduced compared to the unmodified fiber.

15 36. The modified adenovirus fiber of claim 34, wherein the fiber knob is replaced with fiber knobs from an adenovirus that does not interact with CAR.

37. The modified adenovirus fiber of claim 36, wherein the adenovirus fiber knob that does not interact with CAR is Ad3 fiber knob,
20 Ad41 short fiber knob, or Ad35 fiber knob.

38. The modified adenovirus fiber of claim 34, wherein fiber knob mutations are mutations in the AB loop or CD loop.

39. The modified adenovirus fiber of claim 38, wherein fiber knob mutations are mutations in the AB loop or CD loop selected from
25 KO1 and KO12.

40. A modified adenovirus fiber, comprising a fiber protein, wherein:

the unmodified fiber binds the Coxsackie-Adenovirus Receptor (CAR);

-113-

the fiber protein comprises a modification to the fiber protein shaft such that binding of the modified fiber to CAR is substantially reduced or eliminated;

the modified fiber comprises repeats corresponding to the third
5 repeat and the last full repeat; and
at least one repeat of the fiber shaft is deleted.

41. The modified adenovirus fiber of claim 40, wherein the repeats corresponding to repeats 4-17 are deleted.

42. The modified adenovirus fiber of claim 40 or claim 41, wherein
10 the fiber is from a serotype C adenovirus.

43. The modified adenovirus fiber of claim 42, wherein the serotype C adenovirus is Ad2 or Ad5.

44. The modified adenovirus fiber of claim 43, wherein the amino acids corresponding to positions 95-316 are deleted.

15 45. The modified adenovirus fiber of any of claims claim 1-39, wherein the fiber protein is from a serotype A, B, C or F adenovirus; and at least one amino acid corresponding to the consensus repeat sequence as set forth in SEQ ID No. 49 is modified in the repeat corresponding to either the third repeat or the last full repeat.

20 46. A nucleic acid molecule, comprising a sequence of nucleotides that encodes a modified fiber of any of claims 1-45.

47. The nucleic acid molecule of claim 46 that comprises a vector.

25 48. The nucleic acid molecule of claim 46 or claim 47 that comprises heterologous nucleic acid encoding a gene product.

49. The nucleic acid molecule of any of claims 46-48 that is an adenovirus vector.

50. The nucleic acid molecule of claim 49 that is an adenoviral vector from a serotype C adenovirus.

-114-

51. The nucleic acid molecule of claim 49 or claim 50, wherein the heterologous nucleic acid encodes a therapeutic product.

52. The nucleic acid molecule of any of claims 46-51 that is an early generation adenoviral vector, a gutless adenoviral vector or a replication-conditional adenoviral vector.

53. The nucleic acid molecule of claim 52, wherein the replication-conditional adenoviral vector is an oncolytic adenoviral vector.

54. A cell, comprising the nucleic acid of any of claims claim 46-53.

55. The cell of claim 54 that is a eukaryotic cell.

56. The cell of claim 54 that is a prokaryotic cell.

57. A cell of claim 54 that is in a packaging cell line.

58. An adenovirus particle, comprising the modified fiber of any of claims 1-45.

59. The adenovirus particle of claim 58, wherein the capsid further comprises a penton modification.

60. The adenovirus particle of claim 58 or claim 59, wherein the modified fiber includes an N-terminal portion from a fiber of a serotype C Ad virus, wherein the N-terminal portion is sufficient to increase incorporation into the particle compared to in its absence.

61. The adenovirus particle of any of claims 58-60, that comprises a modified serotype C genome, wherein the N-terminal portion of the modified fiber comprises at least the N-terminal 15, 16 or 17 amino acids of a serotype C fiber.

62. The particle of claim 61 wherein the serotype C virus is an Ad2 or Ad5 virus.

63. The adenoviral particle of any of claims 58-62 that further comprises a targeting ligand in the capsid.

-115-

64. The adenovirus particle of any of claims 58-63 further, comprising a heterologous nucleic acid in the genome thereof.

65. The adenovirus particle of claim 64, wherein the heterologous nucleic acid encodes a therapeutically effective product.

5 66. The adenoviral particle of any of claims 58-65 that includes a modification to the capsid whereby binding of the viral particle to HSP is altered compared to a particle that expresses an unmodified capsid.

67. The adenoviral particle of claim 66, wherein the capsid modification that alters HSP binding is in the fiber.

10 68. An adenoviral particle of any of claims 58-67, comprising a mutation in the α_v integrin-binding region of the capsid, whereby binding to the integrin is eliminated or reduced.

15 69. The adenoviral particle of any of claims 58-68, wherein the fiber further comprises a modification in the fiber knob to further reduce any CAR binding.

70. The adenoviral particle of claim 69, wherein the fiber knob modification is in the AB loop or CD loop.

71. The adenoviral particle of claim 70, wherein the fiber knob modification is selected from the group consisting of KO1 and KO12.

20 72. A composition formulated for administration to a subject, comprising the adenovirus particle of any of claims 58-71.

73. A method of detargeting an adenoviral vector, comprising reducing or eliminating the binding of an adenoviral particle to CAR by producing an adenoviral particle that expresses a modified fiber of any of
25 claims 1-45.

74. The method of claim 73, wherein the modified fiber increases the binding to the particular cell type compared to the unmodified fiber.

-116-

75. The method of claim 73, wherein the modified fiber comprises at least two modifications such that the binding to a selected cell type is increased relative to the unmodified fiber.

76. The method of claim 75, wherein the second modification
5 comprises the addition of a targeting ligand in the capsid of the adenoviral particle.

77. The method of claim 75, wherein the second modification comprises the replacement of the fiber knob or a portion thereof.

78. A method, comprising introducing an adenoviral particle of
10 any of claims 58-71 into cells; and introducing the cells into a subject.

79. The method of claim 78, wherein the cells are immune cells or fibroblasts.